Amendments to the Specification

Please amend the specification as follows:

Amend the paragraph beginning on page 2, line 26 as follows:

In one embodiment, an AS3 nucleic acid molecule of the invention is at least 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 98%, or more identical to the nucleotide sequence (e.g., to the entire length of the nucleotide sequence) shown in SEQ ID NO:1 or 3~~ or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____, or a complement thereof~~.

Amend the paragraph beginning on page 3, line 4 as follows:

In another embodiment, an AS3 nucleic acid molecule includes a nucleotide sequence encoding a protein having an amino acid sequence sufficiently homologous to the amino acid sequence of SEQ ID NO:2~~ or an amino acid sequence encoded by the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~. In a preferred embodiment, an AS3 nucleic acid molecule includes a nucleotide sequence encoding a protein having an amino acid sequence at least 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 98% or more homologous to the entire length of the amino acid sequence of SEQ ID NO:2~~ or the amino acid sequence encoded by the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~. In a related embodiment, the nucleic acid encodes a polypeptide fragment having at least 1391 amino acid residues of the amino acid sequence shown in SEQ ID NO: 2.

Amend the paragraph beginning on page 3, line 15 as follows:

In another preferred embodiment, an isolated nucleic acid molecule encodes the amino acid sequence of human AS3. In yet another preferred embodiment, the nucleic acid molecule includes a nucleotide sequence encoding a protein having the amino acid sequence of SEQ ID NO:2~~ or the amino acid sequence encoded by the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~.

Amend the paragraph beginning on page 3, line 20 as follows:

Another embodiment of the invention features nucleic acid molecules, preferably AS3 nucleic acid molecules, which specifically detect AS3 nucleic acid molecules relative to nucleic acid molecules encoding non-AS3 proteins. For example, in one embodiment, such a nucleic acid molecule is at least 100-250, 250-300, 300-350, 350-400, 400-450, 450-500, 500-550, or 550-600 or more nucleotides in length and hybridizes under stringent conditions to a nucleic acid molecule comprising the nucleotide sequence shown in SEQ ID NO:1, ~~the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, or a complement thereof. In a related embodiment, the nucleic acid molecule can further contain a nucleotide sequence encoding a heterologous polypeptide.

Amend the paragraph beginning on page 4, line 1 as follows:

In other preferred embodiments, the nucleic acid molecule encodes a naturally occurring allelic variant of a polypeptide comprising the amino acid sequence of SEQ ID NO:2~~ or an amino acid sequence encoded by the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, wherein the nucleic acid molecule hybridizes to a nucleic acid molecule comprising SEQ ID NO:1 or 3 under stringent conditions.

Amend the paragraph beginning on page 4, line 17 as follows:

Another aspect of this invention features isolated or recombinant AS3 proteins and polypeptides. In one embodiment, the isolated protein, preferably an AS3 protein has an amino acid sequence at least about 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 98% or more homologous to the amino acid sequence of SEQ ID NO:2~~ or the amino acid sequence encoded by the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~.

Amend the paragraph beginning on page 4, line 23 as follows:

In another embodiment, the invention features fragments of the AS3 protein having the amino acid sequence of SEQ ID NO:2, wherein the fragment comprises at least 15 amino acids (e.g., contiguous amino acids) of the amino acid sequence of SEQ ID NO:2~~ or an amino acid sequence encoded by the DNA insert of the plasmid deposited with the ATCC as Accession Number _____~~. In another embodiment, the protein, preferably an AS3 protein, has the amino acid sequence of SEQ ID NO:2, respectively.

Amend the paragraph beginning on page 13, line 30 as follows:

The nucleotide sequence of the isolated human AS3 cDNA and the predicted amino acid sequence of the human AS3 polypeptide are shown in Figure 1 and in SEQ ID NOs:1 and 2, respectively. ~~A plasmid containing the nucleotide sequence encoding human AS3 was deposited with the American Type Culture Collection (ATCC), 10801 University Boulevard, Manassas, VA 20110-2209, on _____ and assigned Accession Number _____. This deposit will be maintained under the terms of the Budapest Treaty on the International Recognition of the Deposit of Microorganisms for the Purposes of Patent Procedure. This deposit was made merely as a convenience for those of skill in the art and is not an admission that a deposit is required under 35 U.S.C. §112.~~

Amend the paragraph beginning on page 15, line 5 as follows:

A nucleic acid molecule of the present invention, e.g., a nucleic acid molecule having the nucleotide sequence of SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, or a portion thereof, can be isolated using standard molecular biology techniques and the sequence information provided herein. Using all or portion of the nucleic acid sequence of SEQ ID NO:1 or 3, ~~or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, as a hybridization probe, AS3 nucleic acid molecules can be isolated using standard hybridization and cloning techniques (e.g., as described in Sambrook, J., Fritsh, E. F., and Maniatis, T. *Molecular Cloning: A Laboratory Manual. 2nd, ed., Cold Spring Harbor Laboratory,* Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, 1989). In a related embodiment, the invention features an AS3 nucleic acid molecule having the sequence of SEQ ID NO: 4 (see Fig. 6) which is identical to the AS3 sequence provided in SEQ ID NO:1 (see Fig. 1) except for an additional 84 base pairs at the 5' end of the molecule. One skilled in the art would recognize that this additional untranslated 5' sequence may indicate that an alternative start site for the beginning of transcription of the AS3 mRNA molecule may exist.

Amend the paragraph beginning on page 15, line 21 as follows:

Moreover, a nucleic acid molecule encompassing all or a portion of SEQ ID NO:1 or 3, ~~or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~ can be isolated by the polymerase chain reaction (PCR) using synthetic oligonucleotide primers designed based upon the sequence of SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~.

Amend the paragraph beginning on page 16, line 8 as follows:

In another preferred embodiment, an isolated nucleic acid molecule of the invention comprises a nucleic acid molecule which is a complement of the nucleotide sequence shown in SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, or a portion of any of these nucleotide sequences. A

nucleic acid molecule which is complementary to the nucleotide sequence shown in SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, is one which is sufficiently complementary to the nucleotide sequence shown in SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, such that it can hybridize to the nucleotide sequence shown in SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, thereby forming a stable duplex.

Amend the paragraph beginning on page 16, line 20 as follows:

In still another preferred embodiment, an isolated nucleic acid molecule of the present invention comprises a nucleotide sequence which is at least about 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 98% or more homologous to the entire length of the nucleotide sequence shown in SEQ ID NO:1 or 3~~, or the entire length of the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, or a portion of any of these nucleotide sequences.

Amend the paragraph beginning on page 16, line 26 as follows:

Moreover, the nucleic acid molecule of the invention can comprise only a portion of the nucleic acid sequence of SEQ ID NO:1 or 3, ~~or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____,~~ for example, a fragment which can be used as a probe or primer or a fragment encoding a portion of an AS3 protein, e.g., a biologically active portion of an AS3 protein. The nucleotide sequence determined from the cloning of the AS3 gene allows for the generation of probes and primers designed for use in identifying and/or cloning other AS3 family members, as well as AS3 homologues from other species. The probe/primer typically comprises substantially purified oligonucleotide. The oligonucleotide typically comprises a region of nucleotide sequence that hybridizes under stringent conditions to at least about 12 or 15, preferably about 20 or 25, more preferably about 30, 35, 40, 45, 50, 55, 60, 65, or 75 consecutive nucleotides of a sense sequence of SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, of an anti-sense sequence of SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~,

or of a naturally occurring allelic variant or mutant of SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~. In an exemplary embodiment, a nucleic acid molecule of the present invention comprises a nucleotide sequence which is greater than 100-150, 150-200, 200-250, 250-300, 300-350, 350-400, 400-450, 450-500, 500-550, 550-600, 600-650, 650-700, 700-750, 750-800, 800-850, 850-900, 900-950, 950-1000, or more nucleotides in length and hybridizes under stringent hybridization conditions to a nucleic acid molecule of SEQ ID NO: 1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~.

Amend the paragraph beginning on page 17, line 26 as follows:

A nucleic acid fragment encoding a "biologically active portion of an AS3 protein" can be prepared by isolating a portion of the nucleotide sequence of SEQ ID NO: 1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, which encodes a polypeptide having an AS3 biological activity (the biological activities of the AS3 proteins are described herein), expressing the encoded portion of the AS3 protein (e.g., by recombinant expression *in vitro*) and assessing the activity of the encoded portion of the AS3 protein.

Amend the paragraph beginning on page 18, line 1 as follows:

The invention further encompasses nucleic acid molecules that differ from the nucleotide sequence shown in SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, due to degeneracy of the genetic code and thus encode the same AS3 proteins as those encoded by the nucleotide sequence shown in SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~. In another embodiment, an isolated nucleic acid molecule of the invention has a nucleotide sequence encoding a protein having an amino acid sequence shown in SEQ ID NO:2.

Amend the paragraph beginning on page 18, line 9 as follows:

In addition to the AS3 nucleotide sequences shown in SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession~~

~~Number _____~~, it will be appreciated by those skilled in the art that DNA sequence polymorphisms that lead to changes in the amino acid sequences of the AS3 proteins may exist within a population (e.g., the human population). Such genetic polymorphism in the AS3 genes may exist among individuals within a population due to natural allelic variation. As used herein, the terms "gene" and "recombinant gene" refer to nucleic acid molecules which include an open reading frame encoding an AS3 protein, preferably a mammalian AS3 protein, and can further include non-coding regulatory sequences, and introns.

Amend the paragraph beginning on page 19, line 5 as follows:

Moreover, nucleic acid molecules encoding other AS3 family members and, thus, which have a nucleotide sequence which differs from the AS3 sequences of SEQ ID NO:1 or 3, ~~or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~ are intended to be within the scope of the invention. For example, another AS3 cDNA can be identified based on the nucleotide sequence of human AS3. Moreover, nucleic acid molecules encoding AS3 proteins from different species, and which, thus, have a nucleotide sequence which differs from the AS3 sequences of SEQ ID NO:1 or 3, ~~or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~ are intended to be within the scope of the invention. For example, a mouse AS3 cDNA can be identified based on the nucleotide sequence of a human AS3.

Amend the paragraph beginning on page 19, line 22 as follows:

Accordingly, in another embodiment, an isolated nucleic acid molecule of the invention is at least 15, 20, 25, 30 or more nucleotides in length and hybridizes under stringent conditions to the nucleic acid molecule comprising the nucleotide sequence of SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~. In other embodiment, the nucleic acid is at least 30, 50, 100, 150, 200, 250, 253, 300, 350, 400, 450, 500, 550, 600, 650, 700, 750, 800, 850, 900, or 950 nucleotides in length. As used herein, the term "hybridizes under stringent conditions" is intended to describe conditions for hybridization and washing under which nucleotide sequences at least 60% homologous to each other typically remain hybridized to each other. Preferably, the conditions are such that sequences at least about 70%, more preferably at least about 80%, even more

preferably at least about 85% or 90% homologous to each other typically remain hybridized to each other. Such stringent conditions are known to those skilled in the art and can be found in *Current Protocols in Molecular Biology*, John Wiley & Sons, N.Y. (1989), 6.3.1-6.3.6. A preferred, non-limiting example of stringent hybridization conditions are hybridization in 6X sodium chloride/sodium citrate (SSC) at about 45C, followed by one or more washes in 0.2 X SSC, 0.1% SDS at 50°C, preferably at 55°C, more preferably at 60°C, and even more preferably at 65°C. Preferably, an isolated nucleic acid molecule of the invention that hybridizes under stringent conditions to the sequence of SEQ ID NO:1 or 3 corresponds to a naturally-occurring nucleic acid molecule. As used herein, a "naturally-occurring" nucleic acid molecule refers to an RNA or DNA molecule having a nucleotide sequence that occurs in nature (e.g., encodes a natural protein).

Amend the paragraph beginning on page 20, line 11 as follows:

In addition to naturally-occurring allelic variants of the AS3 sequences that may exist in the population, the skilled artisan will further appreciate that changes can be introduced by mutation into the nucleotide sequences of SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, thereby leading to changes in the amino acid sequence of the encoded AS3 proteins, without altering the functional ability of the AS3 proteins. For example, nucleotide substitutions leading to amino acid substitutions at "non-essential" amino acid residues can be made in the sequence of SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~. A "non-essential" amino acid residue is a residue that can be altered from the wild-type sequence of AS3 (e.g., the sequence of SEQ ID NO:2) without altering the biological activity, whereas an "essential" amino acid residue is required for biological activity. For example, amino acid residues that are conserved among the AS3 proteins of the present invention, e.g., those present in the heptad repeat or kinase domains, are predicted to be particularly recalcitrant to alteration. Furthermore, additional amino acid residues that are conserved between the AS3 proteins of the present invention and other members of the ASIC family are not likely to be amenable to alteration.

Amend the paragraph beginning on page 21, line 3 as follows:

An isolated nucleic acid molecule encoding an AS3 protein homologous to the protein of SEQ ID NO:2 can be created by introducing one or more nucleotide substitutions, additions or deletions into the nucleotide sequence of SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, such that one or more amino acid substitutions, additions or deletions are introduced into the encoded protein. Mutations can be introduced into SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~ by standard techniques, such as site-directed mutagenesis and PCR-mediated mutagenesis. Preferably, conservative amino acid substitutions are made at one or more predicted non-essential amino acid residues. A "conservative amino acid substitution" is one in which the amino acid residue is replaced with an amino acid residue having a similar side chain. Families of amino acid residues having similar side chains have been defined in the art. These families include amino acids with basic side chains (e.g., lysine, arginine, histidine), acidic side chains (e.g., aspartic acid, glutamic acid), uncharged polar side chains (e.g., glycine, asparagine, glutamine, serine, threonine, tyrosine, cysteine), nonpolar side chains (e.g., alanine, valine, leucine, isoleucine, proline, phenylalanine, methionine, tryptophan), beta-branched side chains (e.g., threonine, valine, isoleucine) and aromatic side chains (e.g., tyrosine, phenylalanine, tryptophan, histidine). Thus, a predicted nonessential amino acid residue in an AS3 protein is preferably replaced with another amino acid residue from the same side chain family. Alternatively, in another embodiment, mutations can be introduced randomly along all or part of an AS3 coding sequence, such as by saturation mutagenesis, and the resultant mutants can be screened for AS3 biological activity to identify mutants that retain activity. Following mutagenesis of SEQ ID NO: 1 or 3, ~~or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, the encoded protein can be expressed recombinantly and the activity of the protein can be determined.

Amend the paragraph beginning on page 24, line 6 as follows:

In still another embodiment, an antisense nucleic acid of the invention is a ribozyme. Ribozymes are catalytic RNA molecules with ribonuclease activity which are capable of

cleaving a single-stranded nucleic acid, such as an mRNA, to which they have a complementary region. Thus, ribozymes (e.g., hammerhead ribozymes (described in Haselhoff and Gerlach (1988) *Nature* 334:585-591)) can be used to catalytically cleave AS3 mRNA transcripts to thereby inhibit translation of AS3 mRNA. A ribozyme having specificity for an AS3-encoding nucleic acid can be designed based upon the nucleotide sequence of an AS3 cDNA disclosed herein (i.e., SEQ ID NO:1 or 3~~, or the nucleotide sequence of the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~). For example, a derivative of a *Tetrahymena* L-19 IVS RNA can be constructed in which the nucleotide sequence of the active site is complementary to the nucleotide sequence to be cleaved in an AS3-encoding mRNA. See, e.g., Cech et al. U.S. Patent No. 4,987,071; and Cech et al. U.S. Patent No. 5,116,742. Alternatively, AS3 mRNA can be used to select a catalytic RNA having a specific ribonuclease activity from a pool of RNA molecules. See, e.g., Bartel, D. and Szostak, J.W. (1993) *Science* 261:1411-1418.

Amend the paragraph beginning on page 44, line 8 as follows:

A transgenic animal of the invention can be created by introducing an AS3-encoding nucleic acid into the male pronuclei of a fertilized oocyte, e.g., by microinjection, retroviral infection, and allowing the oocyte to develop in a pseudopregnant female foster animal. The AS3 cDNA sequence of SEQ ID NO:1 can be introduced as a transgene into the genome of a non-human animal. Alternatively, a nonhuman homologue of a human AS3 gene, such as a mouse or rat AS3 gene, can be used as a transgene. Alternatively, an AS3 gene homologue, such as another AS3 family member, can be isolated based on hybridization to the AS3 cDNA sequences of SEQ ID NO:1 or 3~~, or the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~(described further in subsection I above) and used as a transgene. Intronic sequences and polyadenylation signals can also be included in the transgene to increase the efficiency of expression of the transgene. A tissue-specific regulatory sequence(s) can be operably linked to an AS3 transgene to direct expression of an AS3 protein to particular cells. Methods for generating transgenic animals via embryo manipulation and microinjection, particularly animals such as mice, have become conventional in the art and are described, for example, in U.S. Patent Nos. 4,736,866 and 4,870,009, both by Leder et al., U.S. Patent No. 4,873,191 by Wagner et al. and in Hogan, B., *Manipulating the Mouse Embryo*, (Cold Spring

Harbor Laboratory Press, Cold Spring Harbor, N.Y., 1986). Similar methods are used for production of other transgenic animals. A transgenic founder animal can be identified based upon the presence of an AS3 transgene in its genome and/or expression of AS3 mRNA in tissues or cells of the animals. A transgenic founder animal can then be used to breed additional animals carrying the transgene. Moreover, transgenic animals carrying a transgene encoding an AS3 protein can further be bred to other transgenic animals carrying other transgenes.

Amend the paragraph beginning on page 59, line 26 as follows:

An exemplary method for detecting the presence or absence of AS3 protein or nucleic acid in a biological sample involves obtaining a biological sample from a test subject and contacting the biological sample with a compound or an agent capable of detecting AS3 protein or nucleic acid (e.g., mRNA, genomic DNA) that encodes AS3 protein such that the presence of AS3 protein or nucleic acid is detected in the biological sample. A preferred agent for detecting AS3 mRNA or genomic DNA is a labeled nucleic acid probe capable of hybridizing to AS3 mRNA or genomic DNA. The nucleic acid probe can be, for example, a full-length AS3 nucleic acid, such as the nucleic acid of SEQ ID NO:1 or 3~~, or the DNA insert of the plasmid deposited with ATCC as Accession Number _____~~, or a portion thereof, such as an oligonucleotide of at least 15, 30, 50, 100, 250 or 500 nucleotides in length and sufficient to specifically hybridize under stringent conditions to AS3 mRNA or genomic DNA. Other suitable probes for use in the diagnostic assays of the invention are described herein.

Replace the pending sequence listing with the enclosed replacement sequence listing, pages 1-26.